

Cervix and Vagina

Introduction

The vagina and cervix are discussed together because neither topic is large enough to have its own lesson. More importantly, when viewed from the perspective of reproduction, the ovary makes the oocyte, and the uterus (with the help of some spermatozoa) fertilizes and implants the embryo, then enables the fetus to develop until birth, when the cervix and vagina serve as the conduit through which baby is birthed. The vagina is more than just a birth canal, and the cervix is more than just the distal end of the uterus. But because the cervix and vagina are “below the uterus,” they can be seen as a single functional unit. However, the endocervical cervix has uterine epithelium but doesn’t respond to estrogen and progesterone the same way that the uterus does, whereas the ectocervical cervix has vaginal epithelium continuous with the epithelium of the length of the vagina. The cervix is the transition point from uterus to vagina.

This lesson is primarily about the cervix, cervical cancer, and cervical cancer screening. Cervical cancer is one of the greatest successes in the history of health care. At least where medical care is accessible (such as in the United States), medical science has been able to nearly eliminate cervical cancer simply through screening—Pap tests and coloscopies. Cervical cancer is caused by HPV infection, which you learned about in Microbiology. Not only are the screening practices so good, but recently, vaccines have been developed that prevent infections by high-risk HPV serotypes. That means **immunity against cancer**. In addition to screening and vaccines, in 2019 medical science further solidified its dominance over cervical cancer with a risk-stratification algorithm (created by the ASCCP—American Society of Colposcopy and Cervical Pathology, which we at OME like to call the American Society for Cervical Cancer Prevention). Given a woman’s medical information, the algorithm produces a numeric value that guides the next step in management of anyone who screens positive. The system is easily amended as data changes and can even introduce new risk mediators, such as getting the HPV vaccine. We at OME do not believe in checklist medicine—making a diagnosis and choosing an intervention is far more complex than running down a list. Except when it isn’t. Treatment of chronic disease should be evidence-based. Cervical cancer now has so much data behind it that an algorithm CAN direct better health care decisions than most providers can do on their own. Not all disease can be managed by an algorithm, but cervical cancer screening can.

The end of the lesson includes some vaginal histology, a rant on medical sexism, and then a few vaginal/vulvar pathologies. The meat of this lesson is about the cervix and cervical cancer.

Cervix Anatomy and Histology

The cervix is the distal end of the uterus and the origin of the proximal vagina. The lumen of the cervix is continuous with the lumen of the uterus, and opens into the lumen of the vagina. The cervical lumen forms somewhat of a tunnel, termed the **endocervical canal**. On the uterine side, at the **internal os**, the lumens of both the uterus and cervix are continuous, as are their epithelia—both the myometrium and endometrium are continuous throughout the uterus and cervix. The cervix **is the uterus**, but it has very different physiology (more below). On the vaginal side of the tunnel, the **external os** marks the transition point between the uterine epithelium (gland-forming endometrium) and the vaginal epithelium. The glandular epithelium (ciliated columnar cells that invaginate into the lamina propria) very abruptly transitions **from simple columnar** (uterine) to **nonkeratinized stratified squamous** (the same kind of epithelium as the oropharyngeal mucosa). Outside of the reproductive years, that transition is located within the endocervical canal. Within reproductive age, that transition point is generally found just distal to the external os.

The cervix is the thing that keeps a developing fetus within the uterus. The cervix must rapidly dilate in order to allow the fetus passage out of mom and into the world as a neonate. We discuss the changes in the uterus in the Pregnancy island at the end of the Reproduction module. We want to stay focused on the cervix and vagina as a gynecological issue, leaving the obstetrics for later.

The **endocervical endometrium does not proliferate** in response to estrogen the way the uterine endometrium does. The thing that protrudes into the vaginal canal and can be visualized during a speculum exam or colposcopy (we'll describe a colposcopy later in this lesson) is the **ectocervix**. When cancer is suspected, the **endocervix** and **ectocervix** are treated differently. Right now, start thinking, "**endocervix gets endometrial cancer** (because it is simple columnar and gland-forming), **ectocervix gets cervical cancer** (because it is nonkeratinized stratified squamous)."

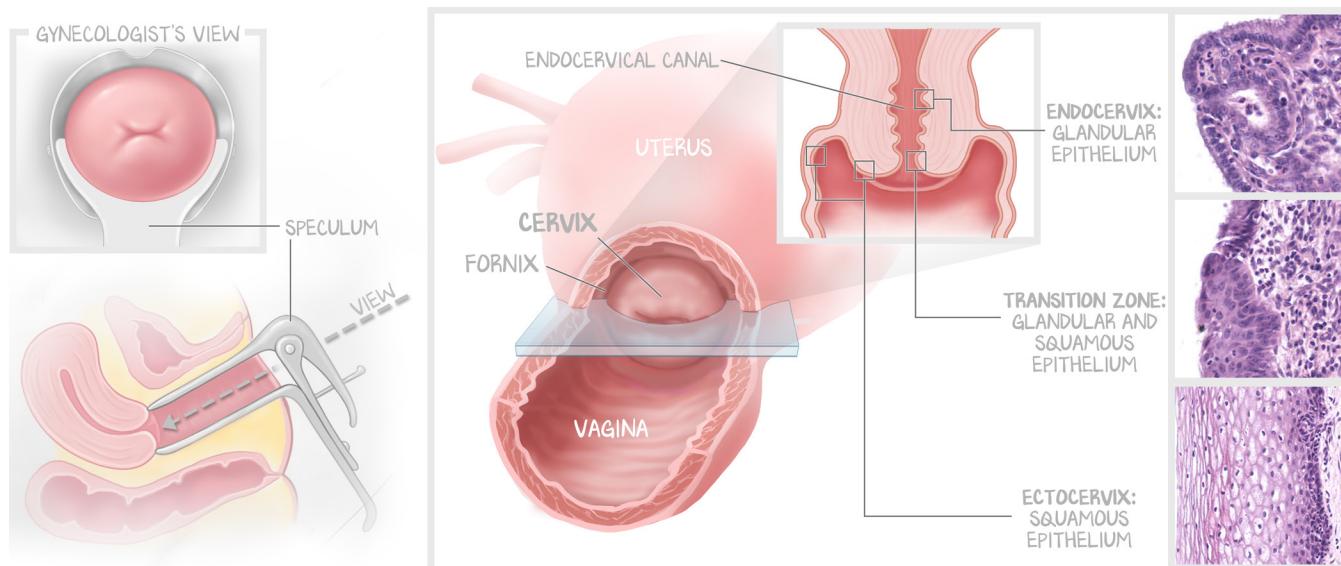


Figure 4.1: Gross Anatomy and Histology of the Cervix

The uterus sits atop the bladder when standing, with the cervix oriented toward the rectum. In the lithotomy position for a speculum exam, the uterus is oriented with the cervix in the view of the speculum. The speculum lifts the walls of the vagina away from the view. The cervix protrudes into the vagina creating the fornix, a divot of vaginal epithelium around the cervix. The ectocervix, the outside of the cervix, inside the vagina, is covered with vaginal epithelium, nonkeratinized stratified squamous epithelium. The endocervix is a simple columnar epithelium that invaginates on its stroma to form glands. The transition zone is the abrupt change from stratified squamous to simple columnar epithelium. During the reproductive years, the transition zone is usually on the ectocervix. In premenarchal girls and postmenopausal women, the transition zone is somewhere in the endocervical canal.

Although the endocervix doesn't have a proliferative or menstrual endometrium, the contents of its ducts change throughout the ovulatory cycle. When in the follicular phase of the ovarian cycle (the proliferative phase of the uterine cycle), the endocervix secretes a harsh fluid that is hazardous to spermatozoa. After ovulation, the progesterone released during the luteal phase of the ovarian cycle changes the endocervical secretions. They become less hazardous and even helpful to the invading spermatozoa. We're also going to discuss the cervix again when we discuss the birthing process.

The History of Cervical Cancer

As you've heard several times throughout our course, "*wherever semen can go, HPV can go.*" Only now, we're adding a modifier—"except when vaccinated against it." Cervical cancer is caused by HPV, so vaccination against HPV is vaccination against cancer. Cervical cancer must still be taught because cervical cancer still

has a high prevalence globally, especially in resource-limited countries. During your career, you will see an end to cervical cancer in the United States—and globally, if the same vaccinations become available.

The improved success of early detection and eradication prior to the development of cancer was achieved through **Pap test** (the former “Pap smear” is no longer used professionally, but may still be used colloquially) screening. The subsequent identification of **human papillomavirus** (HPV; especially serotypes 16 and 18) as the causative agent enabled more advanced screening tools (HPV DNA testing) and, ultimately, the development of the **HPV vaccine**. Originally a bivalent vaccine that only protected against serotypes 16 and 18, newer vaccine formulations include **all the serotypes that can cause cancer** (16, 18, 31, 33, 35, 45, 52, and 58) and some that cause **genital warts** (6 and 8). If medicine is practiced smartly, the only incidence of cervical cancer will be in two populations – the aging population of never-vaccinated patients and those who develop an immunodeficiency (e.g., AIDS). But even then, those women will also need to both contract HPV (which will be unlikely given herd immunity) and avoid cervical cancer screening. We, as a medical community, must ensure that **every child, male or female**, receives the HPV vaccine. With an immune population, HPV will have no reservoir, no hosts to carry it, so it and the cervical cancer it causes will be eradicated. For those who opt out, herd immunity will cover.

In 2019, the US saw over 500,000 total cases of colon cancer, approximately 250,000 of which were in women. By comparison, in the same year, there were only 12,000 cases of cervical cancer. The incidence is currently so low because of screening, and it will fall even further with vaccination. But vigilance is still required. There is no contraindication to the HPV vaccine. **There are two reasons** that boys and girls don’t receive immunity to cervical cancer and genital warts: they aren’t permitted by their parents (an extremely small population of “anti-vaxxers,” whose voices are disproportionately louder than the others in their community) and because **providers of health care don’t offer it**.

The only legitimate risk factor for the acquisition of cervical cancer is **getting HPV**. There are long lists of associated risk factors, like starting to have sex too young, having too many sexual partners (promiscuous sex), not using a condom when engaging in sex, and a history of sexually transmitted infections. They are often taught as if those things were independent risk factors. A girl’s behavior as a teenager doesn’t determine her acquisition of cervical cancer. But those things DO increase the risk of **contracting HPV**. Cigarette smoking increases the risk of cervical cancer by altering secretions. Don’t smoke cigarettes, they’re bad for you. Tell the guy to wrap it up, that’s just pretty smart so you don’t get a different infection, like HIV (that way, you won’t get AIDS). All great advice. ACOG recommends that teenagers **vaccinate against HPV** and **use combined hormone intrauterine contraception**. That means no menses until ready to conceive, avoid pregnancy until ready, and prevention of cervical cancer.

It doesn’t protect the girl from HIV, Hep B, gonorrhea, or chlamydia. So, in addition, coach healthy sex practices. But this lesson is on the history of cervical cancer. **Cervical cancer is HPV**, and **HPV is cervical cancer**. There isn’t another risk factor that matters enough to be mentioned (which is why we listed the ones you will commonly see, so you know why they don’t matter) **if she is vaccinated against HPV**.

HPV and Cervical Cancer

HPV is a DNA virus that is integrated into the host DNA (see Microbiology: Viruses #3: *DNA Viruses*). HPV must infect stem cells at the basement membrane of the stratum basale if it wants to stick around—it’s the only cell layer that proliferates. The HPV that causes cancer can’t get through keratin, so it cannot infect the stratum basale of skin. But HPV is highly tropic for the stratum basale of stratified squamous epithelium. Viral DNA is incorporated into the stem cells’ nuclear material. HPV does not replicate virions in the stem cell, rather HPV induces proliferation of the stem cell. It divides and differentiates a daughter to become a keratinocyte. The HPV then builds viral genome and capsid in the daughter but remains dormant in the stem cell—manipulating genes, stimulating proliferation of the stem cell.

The problem is that HPV infection **accelerates stem cell proliferation**, and proliferation allows for the accumulation of mutations. HPV increases stem cell proliferation and forces stem cells to tolerate mutations through two proteins—E6 and E7. **E6 inhibits p53**, whereas **E7 inhibits Rb**. Rb and p53 are critical checkpoint proteins. Rb is the final molecular signal for the cell to enter S phase, and p53 is the master controller that pauses the cell cycle to enable DNA repair, sends the cell into senescence, or induces apoptosis. When they are inhibited, not only do the basal cells overproliferate, but they also **tolerate the accumulation of mutations**. And according to the stem cell theory of cancer (Inflammation and Neoplasia #7: *Biology of Cancer*), it only takes one malignant cell to develop cancer.

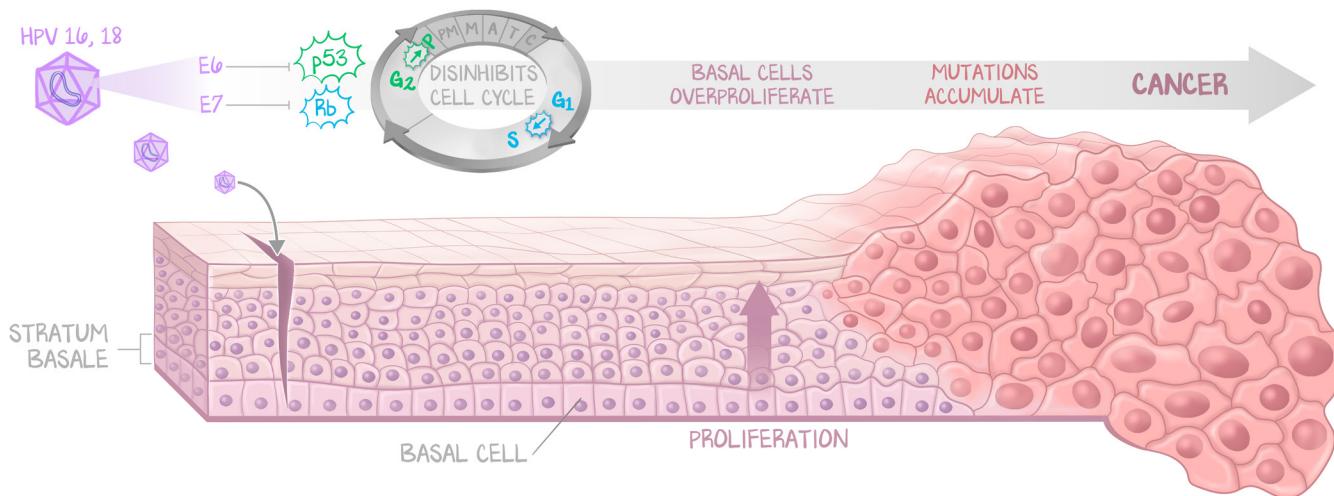


Figure 4.2: HPV Mechanisms

High-risk serotypes of HPV (such as 16 and 18) infect the stem cells of the stratum basale. In that cell, HPV will not produce viral genome or virion. In the cells that are derived from the infected stem cell, it will. But within the infected stem cell of the stratum basale, HPV induces proliferations, which increases the production of squamous cells and therefore makes more virus. HPV achieves these increased proliferations via the proteins E6 (which binds to and inhibits p53) and E7 (which binds to and inhibits Rb). Over time, mutations accumulate due to increased proliferation, eventually resulting in malignancy.

Not every woman with a potentially cancer-causing HPV infection will develop cancer. On average, 50% of HPV infections are cleared within eight months, and 90% of infections are cleared within two years. This means most women who develop cervical cancer were likely exposed multiple times to an HPV-positive partner, even in a monogamous relationship. Even if the virus is eliminated, the proliferation and tolerance of mutations were present. Even a chronic, persistent infection that never clears may not be enough to cause cancer. The accumulation of mutations is random. The immune system may identify the malignant transformation. HPV pumping out proliferations and tolerating mutations merely makes the likelihood of transformation **more likely**. So, the **longer she is infected**, the greater the chance for transformation.

This is why vaccination is so important. If never infected, there is no opportunity for proliferation or the accumulation of mutations.

Cervical Cancer

Patients who are **not screened** will develop a fungating mass which emanates from the cervical os. **Pain with sex** or painless **postcoital bleeding** is a sign of cervical cancer. The diagnosis is made easily by speculum exam, which will demonstrate a large fungating mass. A biopsy will confirm. Staging is based on the local spread (like endometrial cancer, it is a local invasion and not distal metastasis). When HPV infects a non-keratinized stratified squamous epithelium, it causes **squamous cell carcinoma (SCC)**.

In the cervix, which is a nonkeratinized stratified squamous epithelium, HPV induces cervical SCC. Any SCC may demonstrate **keratin whorls** on biopsy. We blew through that illness script because it shouldn't be the one you know and definitely will not be the one you see. **Vaccinate and screen.**

Cervical cancer is most often **squamous cell carcinoma**. Because the endocervix has simple columnar epithelium that invaginates to form glands, and cancer expressing a glandular phenotype is communicated by the word "adeno," whereas cancer of an epithelium is communicated with the word "carcinoma," there can also be **adenocarcinoma** of the endocervix. The Pap test screens for both. But adenocarcinoma is the "other" cancer of the cervix. More often, it is squamous cell. So, unless we specifically call out adenocarcinoma, "cervical cancer" means SCC.

Cytology is obtained from a Pap test, discussed in the next section. **Histology** is obtained from a specialized procedure, a colposcopy, with a needle biopsy, discussed in the section after the next. **Staging** is performed with a physical exam or an MRI. We are not going to teach you staging here, but we will in Clinical Sciences.

Screening Lite: When a Woman Is Never Pap or HPV Positive

This section describes the current screening guidelines for women who are not currently vaccinated against HPV, are not infected with HPV, and have never had an abnormal Pap test. HPV vaccines are new, they aren't recommended for women over 30 years old, and medical science has not yet seen the full impact of vaccination on cervical cancer. You should learn the guidelines in this section as the "old way" despite their currency in clinical practice. Expect to see recommendations change as the impact of vaccination is more fully realized (aka, see the next section).

Screening is performed with Pap tests with or without HPV testing. Start Pap tests without HPV testing at age **21 and repeat every three years**. Start Pap tests with HPV testing (termed **co-testing**) at age **30 and repeat every five years**. As long as all screening is negative, screening can **stop at age 65**.

Do NOT start Pap tests before age 21. Screening was previously recommended to start after the onset of sexual activity. Many girls test positive for either HPV or the cellular atypia typical of an HPV infection, and subsequently clear the virus and have no evidence of pathology or increased risk of cancer later in life. Therefore, the recommendation is now to start screening with Pap tests at age 21.

Do NOT test for HPV until age 30. The highest incidence of HPV infections is in those aged 20–24, more or less in line with increased sexual activity. Almost all of these infections are cleared without an increased risk of cervical cancer. But if a woman has persistent HPV into her 30s, then she is being reinoculated regularly (her sex partner has HPV), or she has a chronic infection. If she didn't already clear an HPV infection, then she won't clear the HPV infection, so she needs more frequent evaluations because her risk of cancer is much higher.

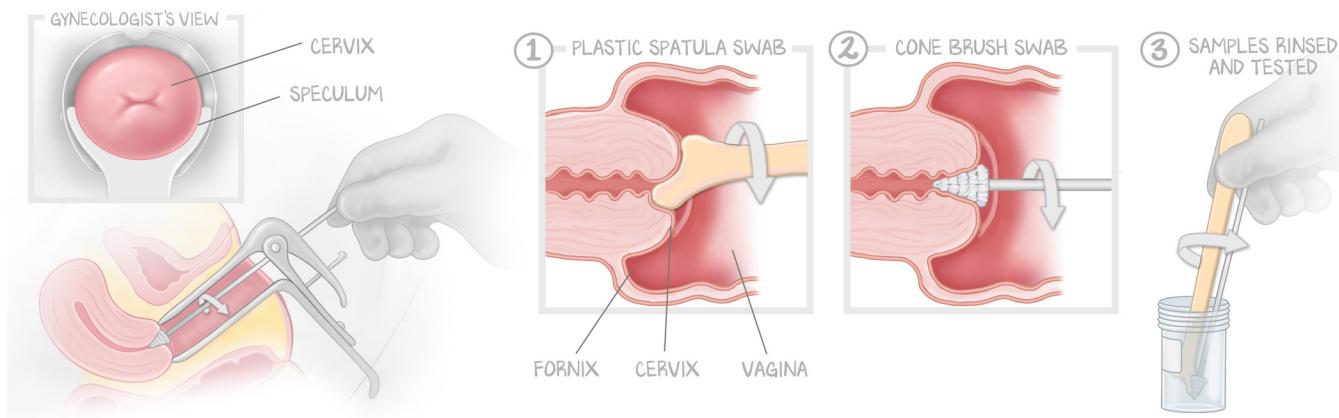
Screening with a Pap test too early (before age 21) or testing for HPV serotypes too early (before age 30) results in too many **unnecessary procedures**. It isn't that the tests are false positives (there is atypia or the virus is present), but a "positive" result means she gets moved along the treatment protocol toward more invasive and painful procedures without any benefit. Coloscopies are painful, and excisional procedures can compromise fertility. The goal is to reduce unnecessary procedures while still reducing cervical cancer.

For **immunocompromised** women, including anyone with HIV, screening starts at the earlier of either the age of sexual initiation or age 21 and continues throughout their life because these women are much more likely to develop cervical cancer.

DO	DON'T
Start screening at age 21 with Pap test q3 years.	Start Pap test screening before age 21.
Start co-testing at age 30 with Pap and HPV tests q5 years.	Start co-testing (Pap and HPV) before age 30. Continue screening past 65 .
Discontinue screening at 65 when adequate screening has been performed.	Continue screening if someone has had her cervix surgically removed (hysterectomy).

Table 4.1

A **Pap test** (named for its inventor, Dr. Georgios Papanikolaou, but shortened for ease of pronunciation) is the first screening tool. The general terminology is **cytology**, but almost everyone in health care uses the term “Pap smear” despite “Pap test” being the name preferred by gynecological societies. The Pap test is performed during a speculum exam. An ectocervix-shaped spatula retrieves a sample from the ectocervix. A cone-shaped brush is introduced into the endocervical canal through the external os. It is crucial to get a good sampling of both.

**Figure 4.3: Acquiring Samples for the Pap Test**

In position for the speculum exam, the woman on her back, the cervix is in direct view, as seen through the speculum. The goal of the Pap test is to acquire samples from the transition zone. This cannot be identified with the naked eye, so two instruments are used. The first is a plastic spatula designed to scrape the ectocervix and get the outer edge of the endocervix. The second is a cone-shaped brush that is introduced into the endocervical canal, targeting the endocervix. The two samples are then taken for testing.

The sample is then examined under a microscope. Because this is performed during a speculum exam, the provider has already visually confirmed that there is no obvious lesion. Thus, we're just talking about screening. Pap test results fall on a spectrum, from no signs of anything wrong to full-blown cancer. There are generally five outcomes of a Pap test: normal, atypical squamous cells of undetermined significance (ASCUS), low-grade squamous intraepithelial lesions (L-SIL), high-grade squamous intraepithelial lesions (H-SIL), and carcinoma. The L-SIL and H-SIL Pap test results have the same name as but are different from the histologic diagnosis made by biopsy, which we discuss below.

Atypical squamous cells of undetermined significance are cells that don't belong (atypical) but don't have obvious features of cancer. From ASCUS, there is an escalation to L-SIL and then H-SIL, depending on how many atypical cells there are and how unusual their behavior is. This means that the test isn't positive or negative, but rather an amalgamation of features that the pathologist uses to

determine how risky the abnormalities are. You will not have to decide the level of atypia and make a diagnosis of ASCUS, L-SIL, H-SIL, or obvious carcinoma, but you should know what to do with the results. What you are about to see should feel extremely foreign.

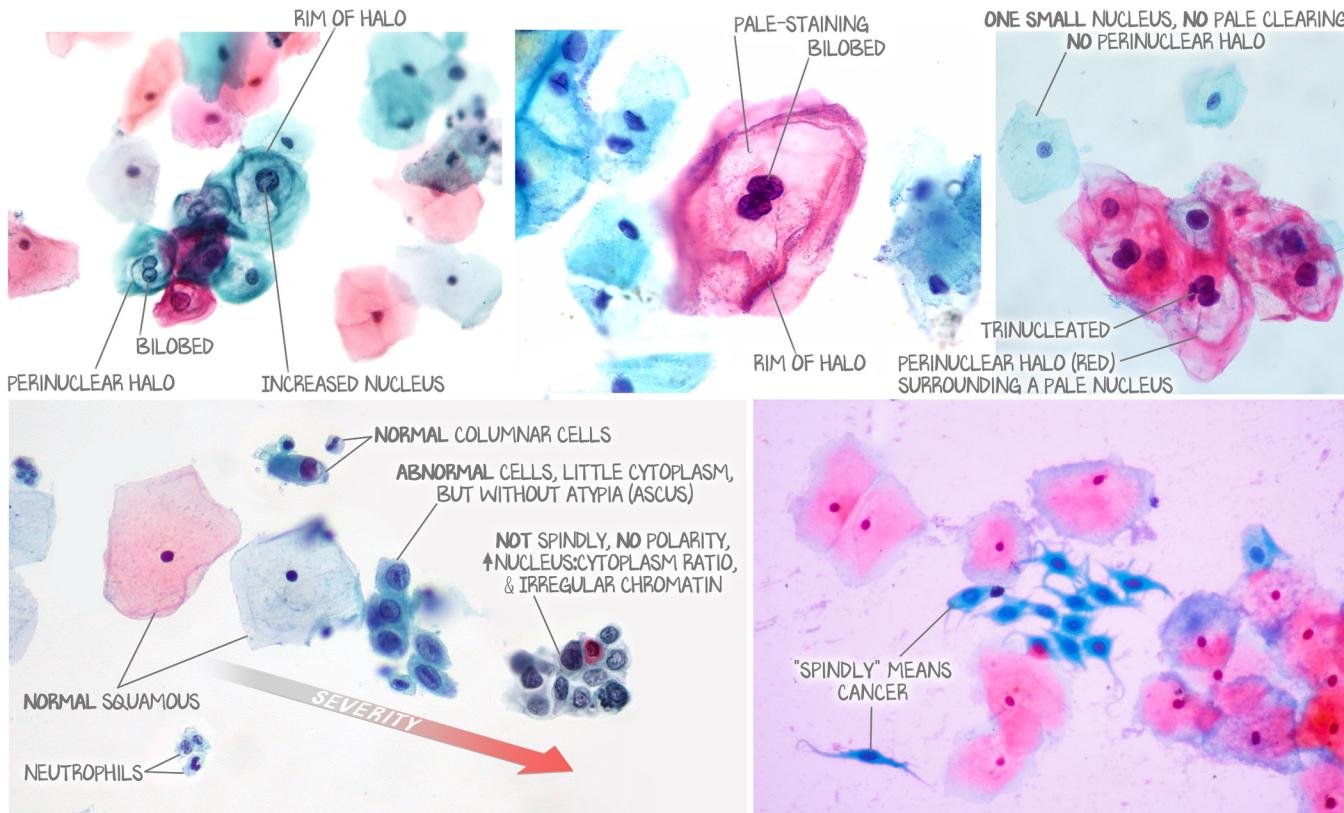


Figure 4.4: Pap Test

There should be two cell types: squamous epithelial cells from the ectocervix and glandular cells from the endocervix. A normal squamous cell will be polygonal with some folds and circular-ish nuclei. They have a very low nucleus-to-cytoplasm ratio and no visible nuclear membrane. It doesn't matter what color they are on this test, as the color is merely a product of the phase of the menstrual cycle or the presence of hormonal contraceptives. But the cells should be uniformly colored—red cells are red, blue cells are blue, etc. The ciliated columnar cells should be just that—ciliated columnar cells with cilia at one side of the cell and the nucleus at the other. When they appear as clusters, they will have clean edges where they abut one another. This is not a detailed lesson on reading Pap tests. We just want to show you what normal vs. bad looks like. Koilocytes are a sign of HPV infection. They have a lighter nucleus with an obvious perinuclear halo and are often binuclear. Dysplasia is detected by assessing the nucleus-to-cytoplasm ratio and the presence of a conspicuous nuclear membrane.

Screening in Depth: When a Woman Is Ever Pap- or HPV-Positive

What the ASCCP did in 2019 was very smart—they built a system that determines the best next step based on the calculated risk of carcinoma in situ of the ectocervix based on the information at hand. If the Pap test is normal, then this system isn't needed, and you just keep screening (Pap test q3y until age 30, then co-testing q5y until age 65). If the Pap test is ever abnormal, feed the available information into the guideline-based algorithm. In goes the information, out comes a number. That number represents the patient's risk of cancer. There are six **clinical action thresholds**, each an escalation of the one before it, that tell you how to manage this patient. If her risk of cancer crosses a clinical action threshold, your next intervention is whatever is above that threshold. This generates **equal management for equal risk**—the components that generated the risk value don't matter, only what the calculated value is. As a miniature example, a co-testing result of HPV-positive with ASCUS cytology has the same risk as L-SIL cytology results without HPV testing. The information you feed it doesn't matter, only the calculated risk.

This system also enables future research to augment the risk stratification and allows the history of HPV vaccination to influence decision making. Factors that increase calculated risk include age, history of (or currently more severe) cytology findings, HPV-positive history and/or current cytologic evidence of HPV infection. Factors that decrease risk include benign cytology findings, no history of HPV, HPV vaccination, and anti-HPV antibody titers. There are more than just these features (including follow-up cytology or histology findings), but we would rather you have the gist of what this clinical tool does than learn every element of how the algorithm produces the risk value—because the algorithm will do it for you.

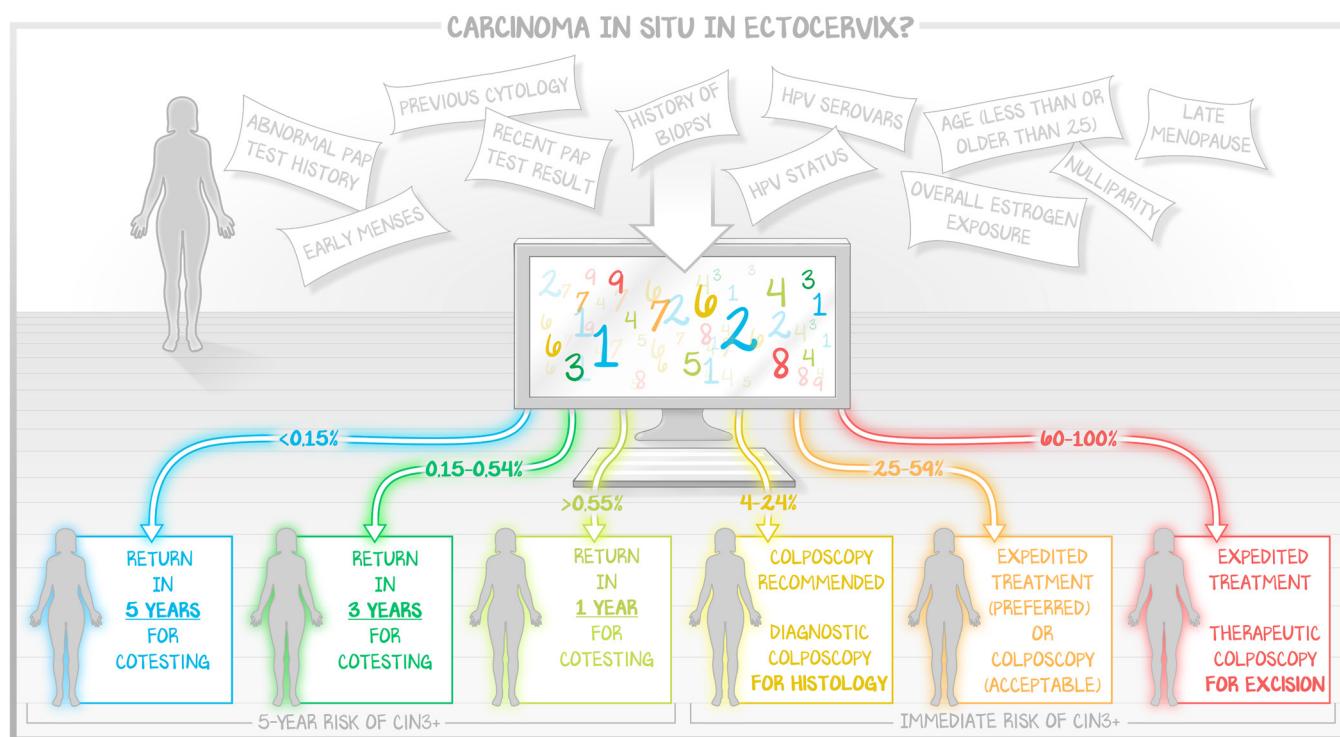


Figure 4.5: New Guidelines—Risk Score to Next Intervention

Technically, when < 4% the algorithm calculates the 5-year risk of developing CIN 3+ and determines when a follow up screening is required. Technically, when > 4% the algorithm calculates the risk of CIN 3+ in this woman right now, and determines whether a diagnostic procedure (colposcopy with biopsy) or a therapeutic procedure (colposcopy with excisional biopsy) is required. But because the algorithm outputs a clinical threshold, you can think of this as a continuum, a spectrum from 0% to 100%, with screening if low risk and treatment if high risk.

Expedited treatment means performing a colposcopy with the intention of excision without first performing a colposcopy to get a biopsy (threshold 60%; don't memorize the thresholds, just get the gist). **Expedited treatment OR colposcopy** is the next category (threshold 25%). The guidelines state that expedited treatment is preferred for this level of risk, but there must be a consideration for age and desired fertility, something you can talk to the patient about. The former guidelines called for no expedited treatment for an abnormal Pap, only a **colposcopy** for inspection and histological evaluation (now threshold 4%). It was a convoluted mess that took age and HPV status into account to determine whether the colposcopy could be avoided, and the woman could simply be re-entered into screening. The new guidelines do that for you. If the risk doesn't reach 4%, the patient is entered into **surveillance**. Surveillance consists of co-testing (Pap and HPV tests), and risk thresholds determine the screening interval—every **1 year** (0.5% threshold), **3 years** (0.15% threshold), or **5 years** (no threshold).

What you do with **follow-up cytology results** is also in the new guidelines, where they go into intense detail that we don't have to look at. Good news: the repeat cytology is tossed back into the algorithm, the new data adjusts her risk, and the new risk value is compared to the same intervention thresholds.

A **colposcopy** is a speculum exam performed with a colposcope—a binocular microscope—that enables better visualization of the cervix. Acetic acid is pipetted onto the cervix. Areas of accelerated mitosis (i.e., where the cells that demonstrated atypia on the Pap test are likely to be coming from) will take up more acetic acid than areas of healthy cervix, so the lesion will appear bright white. The tissue that collects acetic acid is the tissue that you biopsy. **Ectocervical biopsy** takes a sample for histological review. In addition, **endocervical curettage** is performed in nonpregnant patients; for pregnant women without obvious cancer, it's generally better to test after delivery, so as not to risk the pregnancy. The biopsy is used both to confirm that treatment is needed and to assess for malignant transformation (invasion of the basement membrane).

Treatment means either **excision** via loop electrical excision procedure (LEEP) or **ablation** with cryotherapy (cold knife) or a CO₂ laser. **Excision is preferred** because it enables histological review. You can't know how deep a lesion goes if you burn it because you only ablate the surface. With excision, you can be sure you removed the cancer because the excisional biopsy is deep. The risk of infertility increases with excision, which is why the choice is given. A diagnostic colposcopy with acetic acid and biopsy is the same procedure as a therapeutic colposcopy with excisional biopsy—performed in the same room, by the same gynecologist, with the same equipment. The idea is that if there is no obvious lesion, chances are the risk is low, so biopsies are taken. If there is an obvious lesion, chances are the risk is high, so an excision is performed. No need to make the woman go through two colposcopies if she obviously needs an excision procedure.

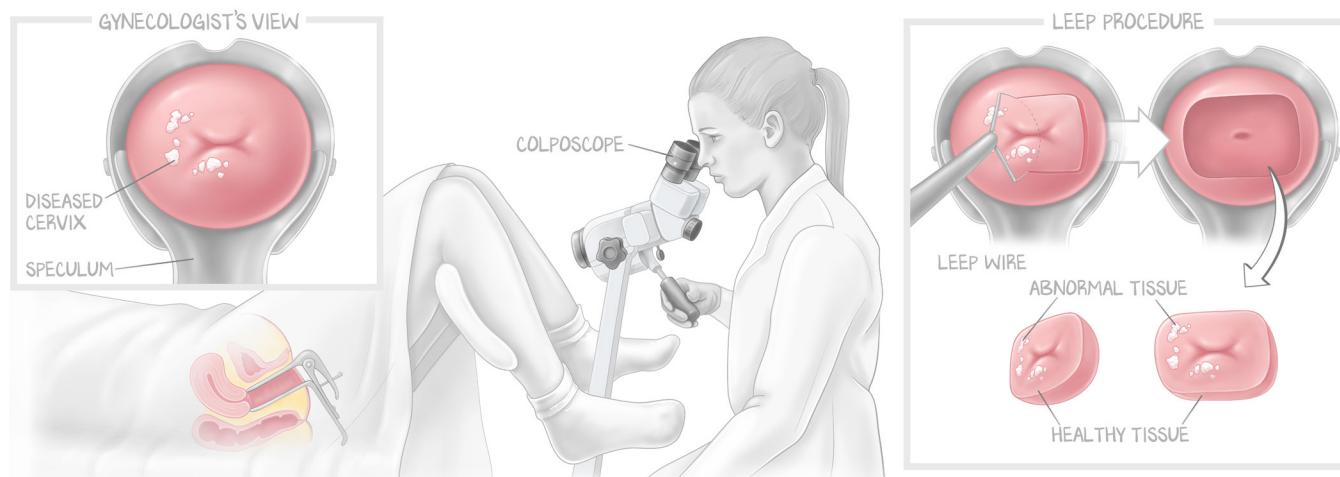


Figure 4.6: Colposcopy and LEEP

A colposcopy is a cervical exam performed using a microscope. For most procedures, the provider looks through the colposcope. New advances in the technique involve using devices that project to television screens or monitors, and don't require the use of a colposcope lens (not depicted). This enables a close inspection of the cervix and placement of acetic acid onto the cervix. Areas that are bright white are either biopsied with a needle (diagnostic colposcopy) or excised with a procedure such as LEEP (therapeutic colposcopy). Other forms of destroying the bad tissue include ablation (burning) rather than excising. Excision is preferred because the provider can inspect the excised lesion for clean margins. Ablation still remains an option, especially if fertility is still desired.

All of the above describes screening for SCC that passes through cervical intraepithelial neoplasia (CIN) with dysplastic squamous cells. Endocervical cancer (uterine epithelium) has a much simpler algorithm. If ever there is cytologic evidence of **atypical glandular cells** (the endometrial cells from the

endometrial “glands”) or **adenocarcinoma in situ** (cancer of the endocervix), the patient is escalated to colposcopy regardless of age, HPV status, or guideline score.

Histology (from a biopsy, not a Pap test) was formerly given a CIN grade from 1 to 3. Often misinterpreted as how much of the epithelium was involved (e.g., CIN 1 stratum basale, CIN 3 full thickness, CIN 2 in-between), it was supposed to determine the level or severity of dysplasia, the risk of progressing on to carcinoma. Answering that question meant more than “how thick the cancer is,” and requires the use of various immunostaining (the figure below).

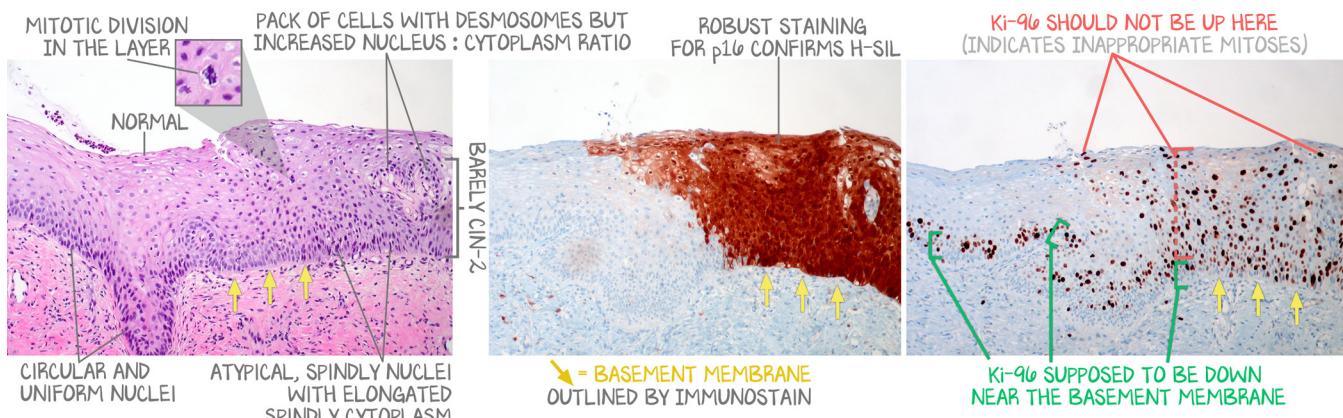


Figure 4.7: Grading Cervical Intraepithelial Neoplasm

(a) Light microscopy with H&E staining demonstrates normal cervical epithelium on the left and spindle-shaped nuclear atypia, increased nucleus-to-cytoplasm ratio, though the entire height epithelium is not involved. (b) When the same sample is stained with p16, the affected regions become more obvious. The lesion is contained by the basement membrane (black arrows), and the brown staining is both the entire height of the epithelium and very dense in the affected regions. At the periphery of the lesion (left side) there is some staining of normal cells—the pale staining, though brown, is an artifact of the procedure. (c) A Ki-96 stain identifies mitosis. The only cells that should be dividing in the cervical epithelium are the stem cells found in the stratum basale. The presence of staining anywhere else indicates a cancerous growth.

Although the CIN methodology provided accurate histology, it had little utility in developing screening and treatment protocols. Pathologists report CIN 1–3. Gynecologists use the **squamous intraepithelial lesion (SIL)** score—**L-SIL** (aka CIN 1) or **H-SIL** (anything more severe than L-SIL)—because **L-SIL does not progress directly to invasive cancer, and typically regresses**. Only a small percentage of L-SIL cases progress on to H-SIL. In contrast, any other SIL is considered **H-SIL** and **premalignant**.

If there is **invasion of the basement membrane**, it is no longer precancerous but rather cervical cancer. Because the cervix comes from the uterus, and uterine cancers grow locally, so too does cervical cancer. Treatment is with excision, chemotherapy, and radiation based on stage. Because we aren’t teaching you staging, we also aren’t teaching you treatment.

Other Cervix-Related Things

Endocervical polyps are common benign exophytic growths that arise within the endocervical canal. They vary from small sessile “bumps” to large polypoid masses that may protrude through the cervical os. Their main significance is that they may be the source of irregular vaginal “spotting” or bleeding that arouses suspicion of some more ominous lesion. Simple curettage or surgical excision is curative. It is the endocervical equivalent of a uterine polyp.

Lactobacillus makes up the majority of the human vaginal microbiome. As the nonkeratinized stratified squamous epithelium terminally differentiates and eventually desquamates (as all stratified squamous epithelium does), the epithelial cells produce a substantial amount of glycogen. This glycogen

is effectively fed to the *Lactobacillus* that lives in the vagina. These bacteria are part of the microbiome that prevents infections by other organisms. *Lactobacillus* species as the dominant organism in the vaginal microbiome seems to be **unique to humans**, in whom *Lactobacillus* makes up more than 70% of the vaginal microbiome. In every other species with a vagina, it makes up less than 1%. *Lactobacillus* species are not very virulent, so if a woman receives antibiotics for bacterial infection, temporary disruption of the vaginal microbiome can lead to opportunistic infections, such as with *Candida albicans*. We cover cervicitis and vaginitis in Clinical Sciences, so we aren't spending long on them here.

Vaginal Anatomy and Histology

The vagina is a muscular tube with one large lumen. Like the uterus and cervix, its upper third is formed by the distal ends of the Müllerian ducts, and so not surprisingly, the vaginal artery is actually the vaginal branch of the uterine artery. Where the cervix protrudes into the vagina, there is a recessed rim called the **fornix**. The vaginal fornix is a ring, so it doesn't have sides. Just like any organ, it can be described by its relative location (the anterior fornix is anterior to the posterior fornix). The only thing that matters about the fornix (never fornices, there is just one) is that the **posterior aspect** of the fornix provides surgical access to the **rectouterine pouch**, where fluid (pus, blood, or just ascites) that shouldn't be in the peritoneal cavity may accumulate. The lower two-thirds of the vagina are formed by the urogenital sinus and are continuous with the upper third. The vascular supply of the lower vagina comes from the **internal pudendal artery** and sometimes the rectal arteries. The epithelium is **nonkeratinized stratified squamous** epithelium, identical to and continuous with the ectocervical mucosa. Just as we saw with the uterine tubes, uterus, and cervix, the vagina has a mucosa (epithelium on a lamina propria), has no submucosa, and is wrapped in muscle.

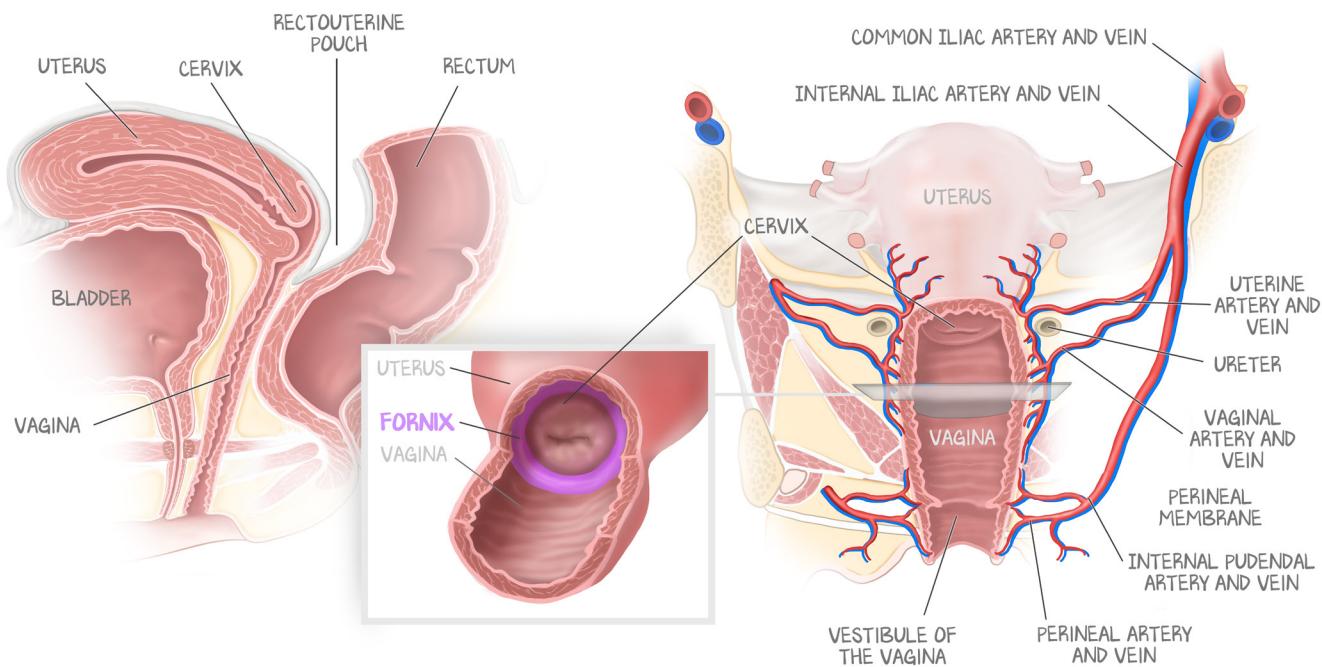


Figure 4.8: Vaginal Anatomy

The vagina is the tube that connects the uterus to the outside world. The uterus lies over the bladder, anterior to the cervix, in the normal upright position. To see the cervix, which protrudes into the vagina, a woman lays on her back in footrests (also called stirrups) to orient the cervix towards the examiner. The vagina is not rigid and has a soft mucosa and floppy walls, much akin to the esophagus. This is why cervical evaluation requires the use of a speculum to maintain visual patency. Unlike the rest of the reproductive organs, the vagina is below the pelvic floor, so there are no ligaments to worry about. The blood supply varies along the length of the vagina, but we want you to learn the internal pudendal artery as its blood supply.

The Hymen and a Rant On Medical Sexism by Dr. Williams

This would be Dr. Williams here writing this section. It baffles me that even prominent textbooks, ones I value and have learned a lot from, still have lines like this: "*In the virgin, the entrance to the vagina may be covered by a mucosal membrane . . . hymen.*" The first problem with that statement is the use of *the virgin*, evaluating a female human (girl or woman) based on her personal history of being penetrated in the vagina by a human penis (the sex act that "counts" as devirginizing her). The second problem is that something other than a male human penis can perforate the hymen instead, making her still a *virgin* human female who has not had her vagina penetrated by a male human penis, but without a hymen. The third problem—and this one isn't as important as the first two, but is illustrative of an author who lacks perspective—is the assumption that the *entrance* to the vagina is the most superficial, closest to the male who wrote the paragraph in the textbook, and not the *exit* of the vagina, the most distal from the ovaries.

I'll explore this a little further. Girls tend to have menses before sex, and as the hymen is a thin membrane (a remnant of the urogenital membrane) that was supposed to go away in the first place, a tampon may be sufficient trauma to eliminate it. Now that the ACOG is recommending hormone intrauterine contraception for girls as young as 13, she is going to have had a vaginal exam and a device placed in her uterus at a young age. Both the exam and the insertion of the device would require the tearing of the hymen. Some girls are going to have their hymen torn the first time they have vaginal intercourse, and in those girls, there may be pain and bleeding the first time they have vaginal intercourse. So, it is possible that a girl who has not yet had vaginal intercourse has a remnant of the urogenital membrane that partially covers the exit to her vagina. Not having one can be explained by normal development, trauma from anything other than sex, a gynecological exam, use of tampons, sexual exploration prior to copulating with the opposite sex, or anything else. There isn't a "normal hymen" or variations of what one could look like. It is a remnant of a membrane present during embryogenesis that doesn't have a role in adult life.

Now, if evaluating a child for sexual abuse, it may be helpful to find an "intact" hymen, suggesting (not ruling out) that her vagina had not been penetrated (by anything). But that finding doesn't mean she wasn't sexually assaulted. Oh, and when performing that evaluation on a sedated child, there's a lot more you should be looking for than just an intact hymen.

There is utility as a provider of health care in knowing whether a girl is having sex or not, knowing whether she is "a virgin." But the question you ask isn't, "are you a virgin?" but rather, "are you sexually active?" This isn't a communication lesson, so how you go about asking an adolescent about her sex practices is more complicated than this, but the point is that sexual activity is more than having her vagina penetrated by a human penis. And *virgin* is a label, a judgment. Her being genetically and phenotypically female provides a clinical utility (which is why I insist on the use of the substantive adjectives male and female, and why you see it in the opening of every clinical vignette) regarding the risk of disease, expected clinical course, development, etc. It's also why I still use substantive adjectives like *diabetic* and *cirrhotic* despite organizations suggesting that "person with diabetes" is preferred. *Diabetic* is one word that conveys immense clinical utility and leaves no room for heuristic error (does "person," "with," or "diabetes" carry the most import?). But the adjective *virgin* conveys absolutely NO clinical utility. And the reason you ask is so that you can provide counseling on safe sex practices and evaluate her readiness to be engaging in sexual activity, need for contraception, risk of contracting certain infections, readiness for getting pregnant and motherhood, etc. Let's not label a person based on her *virginity* having been penetrated in the vagina by a human penis.

Soapbox over. Sort of. **Imperforate hymen** presents with a bulging blue membrane, pelvic pain during menstruation, only the girl has never actually had any bleeding. What did the lower two-thirds of the vagina form from? The urogenital sinus. That thing starts with the cloacal membrane, which gets renamed to the urogenital membrane when the rectum separates from the urogenital sinus. Urogenital . . . membrane? The "opening" of the urogenital sinus starts closed, covered by a membrane. Because urination and defecation are possible in a neonate without a structural pathology, it would

appear that during the natural course of embryogenesis, the urogenital membrane . . . disappears. The covering of the urethra is supposed to go away. That covering that is supposed to go away over the exit of the urethra is the same covering that is supposed to go away over the exit of the vagina. Does that membrane entirely disappear in its entirety in every female birth? Nope. What are the “variations of the hymen” (variations on the remnant of the urogenital membrane in the postnatal female phenotype)? ANYTHING. Because the membrane doesn’t go away all the way, any remnant of that flimsy membrane may persist in any orientation to the vagina. In the case where the **vaginal membrane is fully intact** (the exit to the vagina is completely obscured), the patient is going to develop through childhood normally. When she has her first menses, the blood and endometrium will not be able to exit the vagina. This is going to hurt, come to the attention of medical providers, who would then perforate the **hymen** vaginal membrane to release the blood and endothelium. Oh, also, if any medical provider ever evaluated the child (did a physical exam), they would already know there was no exit to the vagina and could fix it immediately. It is uncommon, easily diagnosed, easily treated, rarely encountered (PubMed shows 253 case reports or series for a whopping 213 patients) . . . but it shows up on licensing exams all the time.

Vulvar/Vaginal Pathologies

Bartholin glands are stimulated by the parasympathetic nervous system and act as part of the female sex act. Their secretions provide lubrication for the act of sex. If they become obstructed, they become a nidus for infection or abscess. Bartholin duct cysts are relatively common, occur at all ages, and result from obstruction of the duct by an inflammatory process. These cysts are usually lined with transitional or squamous epithelium. They can be easily drained or marsupialized (permanently opened) should recurrent infections persist.

Vulvar and vaginal cancers are extremely rare; vulvar cancers account for 4% of gynecologic cancers, and vaginal cancers account for 3%. Thankfully for your learning, the most common cancer phenotype is **squamous cell carcinoma**. The labia minora and vagina are mucocutaneous tissues with nonkeratinized stratified squamous epithelium and are, therefore, vulnerable to HPV infection, which predisposes them to SCC—just as HPV predisposes for cervical, and SCC is just as treatable. No screening method has been developed for vaginal or vulvar cancer because they are so rare.

Diethylstilbestrol (DES) hasn’t been used in the United States since 1971, but it still rears its ugly head on licensing exams. DES is a nonsteroidal estrogen used to prevent miscarriages. As it turns out, it’s also a teratogen that causes reproductive tract abnormalities and developmental abnormalities, and predisposes one to **clear cell adenoma of the vagina**. Those exposed would develop this cancer in the first few years of life. Because no one in this country has received DES for 50 years, you will never see clear cell adenoma of the vagina in real life.

The developmental abnormalities caused by DES include a septate vagina (double vagina) due to the failure of the Müllerian ducts to fuse, much like we saw in the uterus. The vagina is initially covered by **endocervical-type columnar** epithelium. This is typically replaced by squamous epithelium, advancing upward from the urogenital sinus during development. If some patches of columnar epithelium persist, the patient is diagnosed with vaginal **adenosis**, as the epithelium is endocervical, simple columnar that invaginates into glands, thus the “adeno” of vaginal adenosis. It presents clinically as red, granular areas that stand out from the surrounding normal pale-pink vaginal mucosa. In women exposed to DES (none has been in 50 years), adenosis had an incidence of 35% to 90%. But almost no woman has the condition today. There are women alive now who could have been exposed, but as with clear cell adenocarcinoma, the condition would have presented itself by now, as it is a disorder of development in utero.

Embryonal rhabdomyosarcoma, also called sarcoma botryoides, is another uncommon vaginal tumor that shows up on licensing exams. Like clear cell adenocarcinoma, it occurs in infants and girls up to five years old. It is composed of malignant embryonal rhabdomyoblasts that grow with the appearance of a bunch of grapes (hence the designation botryoides).